·Xu et al.

U.S.S.N.:

09/936,588 3/29/02

Page 2

IN THE CLAIMS

Please cancel claims 33-54 and add new claims 55-72.

1-54: (cancelled)

55. (new): A method of reconstituting a target protein from protein fragments in a plant, comprising:

- (a) splitting a gene encoding a target protein into at least two DNA fragments;
- (b) separating the DNA fragments of step (a) to prevent transmission of the gene to other plants; wherein one of the DNA fragments coding for a portion of the target protein is compartmentalized in the nucleus, and the other DNA fragment coding for another portion of the target protein is compartmentalized in the chloroplast;
- (c) expressing the DNA fragments of step (b) within the plant to produce the corresponding fragments of the target protein; and
- (d) reconstituting the target protein from the protein fragments in the plant.
- 56. (new): A method of preventing transmission to a second plant of a gene coding for a target protein in a first plant, comprising:
- (a) splitting the gene encoding the target protein into at least two DNA fragments; and

·Xu et al.

U.S.S.N.: Filed:

09/936,588 3/29/02

Page 3

(b) separating the DNA fragments of step (a) wherein one of the DNA fragments coding for a portion of the target protein is compartmentalized in the nucleus of a host cell in the first plant, and the other DNA fragment coding for another portion of the target protein is compartmentalized in the chloroplasts of the host cell; and

- (c) preventing transmission of the gene coding for the target protein to the second plant .
- 57. (new): A method according to claim 55, wherein at least one of the DNA fragments is fused to a DNA sequence encoding a transit peptide for transport into a chloroplast or nucleus.
- 58. (new): The method of claim 55 or 56, wherein at least one of the DNA fragments is fused to a DNA coding for an intein or portions thereof.
- 59. (new) The method of claim 58, wherein one of the DNA fragments is formed by linking a 5' end of the DNA fragment coding for an N-terminal portion of the target protein to a 3' end of the DNA coding for an N-terminal portion of the intein, and another of the fusion fragments is formed by linking a 5' terminal end of DNA encoding a C-terminal portion of the target protein to the 3' end of DNA coding for a C-terminal portion of the intein.

· Xu et al.

U.S.S.N.:

09/936,588 3/29/02

Filed: Page 4

60. (new): The method of claims 55 or 56 wherein the DNA coding for the target protein is split to form two or more DNA fragments by

means of a DNA coding for one or more affinity domains.

61. (new): The method of claim 60, wherein the affinity domain is

selected from the group consisting of inteins or intein fragments,

leucine zipper and c-Jun/c-Fos.

62. (new): The method of claim 58, in which at least one of the DNA

fragments coding for the target protein is fused to a DNA sequence

encoding a transit peptide such that the protein product of the DNA

fragment is transported into a single compartment where functional

reconstitution can occur.

63. (new): The method of claim 58, wherein reconstitution of the

target protein fragments comprises intein-mediated splicing.

64. (new): The method of claim 58, wherein reconstitution of the

target protein fragments comprises intein-mediated protein

complementation.

65. (new): The method of claim 55, wherein reconstitution of the

target protein fragments comprises protein complementation.

66. (new): The method of claim 65, wherein protein complementation

occurs in the presence of an affinity domain.

· Xu et al.

U.S.S.N.:

09/936,588 3/29/02

Page 5

67. (new): The method of claim 65, wherein protein complementation occurs in the absence of an affinity domain.

- 68. (new): The method of claim 55 or 56, wherein splitting of the gene comprises:
- (a) determining one or more potential split site regions of the target protein; and
- (b) splitting the DNA coding for the target protein at the potential split site region.
- 69. (new): The method of claim 68, wherein the potential split site region of the target protein is determined by analyzing primary amino acid sequence of the target protein for non-conserved regions.
- 70. (new): The method of claim 68, wherein the potential split site region is determined by linker tolerance of linker insertion within the target protein.
- 71. (new): The method of claim 68, wherein the potential split site region is determined by analyzing the structure of the target protein for the presence of flexible loops.
- 72. (new): The method of claim 68, wherein the potential split site region is determined by analyzing the structure of the target protein

Xu et al. U.S.S.N.: Filed:

09/936,588 3/29/02

Page 6

for the presence of amino acid sequence between folding domains of the target protein.